Utilisation of Disease Resistance Genes in Cotton

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Summary

Plants may look simple and defenceless, however on-going genetic research is revealing a complex system that detects harmful organisms and triggers a battery of plant defence responses. We are studying this process of recognition and response in the cotton plant, with the aim of developing molecular tools to ensure that broadly based and effective disease resistance is incorporated into commercial cotton cultivars.

Self-defence in the plant world

Plants share the environment with many micro-organisms, some of which are pathogens with the potential to cause serious harm to the plant. Fortunately, most pathogens do not succeed in injuring the plant, since the plant recognises the organism as a threat and quickly mounts defensive measures. A crucial step that determines a plant's success in fending off an intruder therefore lies in early detection of the pathogen.

Recent research has provided insight into exactly how plants recognise a pathogen. Studies of the DNA of plants have uncovered an extensive collection of genes that encode proteins with the ability to recognise harmful organisms. These disease resistance genes (R genes) are present in hundreds to thousands of copies in the plant genome. Surprisingly, all of the resistance proteins encoded by these R genes have regions of similar structure despite their ability to detect and respond to pathogens as diverse as bacteria, viruses, fungi, nematodes, or even sucking pests such as aphids.

The disease resistance proteins broadly consist of two components. One part is designed to specifically recognise a particular pathogen and not others, a fact that explains the need for such a large collection of resistance genes in each plant. The other part of the protein responds to the recognition of the intruder by sending a signal to the plant cell nucleus. The message reads to quickly turn on the appropriate defence to fend off the attacking pathogen. This series of events is outlined in Figure 1.

Isolation of R gene analogues from cotton

As a result of efforts by plant scientists worldwide, significant progress has been made in the cloning of disease resistance genes from numerous plant species. Furthermore, simpler means of disease resistance gene discovery have arisen from the finding that groups or families of resistance proteins share similar structures regardless of the plant species.

We have taken advantage of this discovery to conduct a search for disease resistance genes in cotton. A powerful technique called the polymerase chain reaction (PCR) was employed to amplify segments of putative disease resistance genes from the cotton genome. The outcome of these experiments was extremely successful, with several groups of genes analogous to

known resistance genes being identified in cotton. Such genes are known as resistance gene analogues or RGAs.

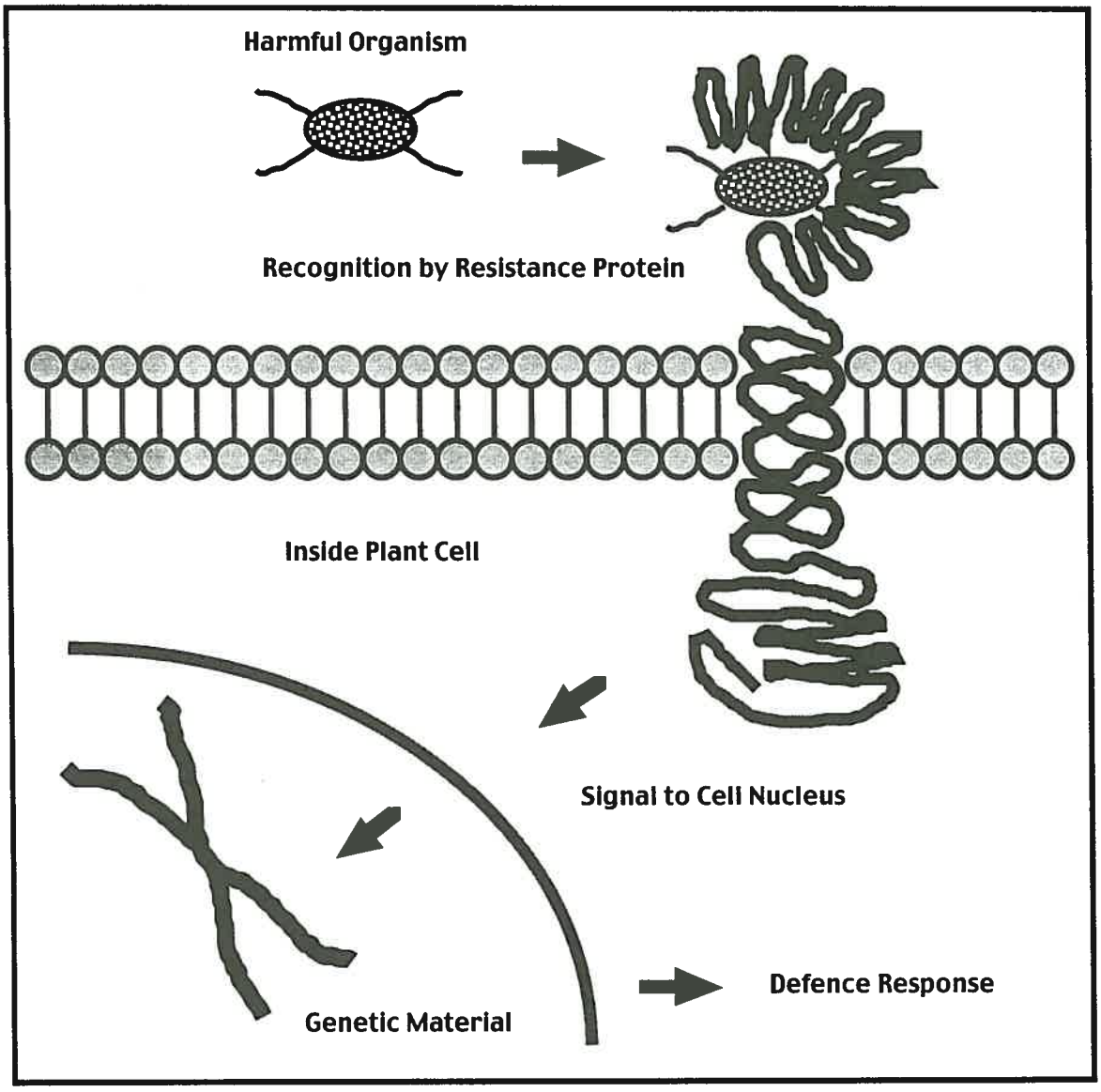

Figure 1. Diagram of a plant disease resistance protein in action. A portion of the protein (black) lies outside the cell and specifically recognises the harmful organism. The remaining portion of the protein (grey) resides inside the cell and communicates a signal to the plant's genetic material, which in turn stimulates a defence response against the invading organism.

Subsequent cloning and characterisation of the cotton RGAs revealed the existence of at least six very different gene families in cotton. Gene copy number was found to be extremely variable between families, with the smallest family apparently present as only a few copies, whereas the largest family appears to exist as hundreds of copies in the cotton genome. Comparative studies using databases of DNA sequence information showed similarities with known resistance genes from plants such as tobacco, flax and the experimental plant *Arabidopsis*. Figure 2 depicts a family tree showing the relationship between members of the

six families of cotton RGAs isolated during the course of our research and resistance genes or RGAs from other plant species.

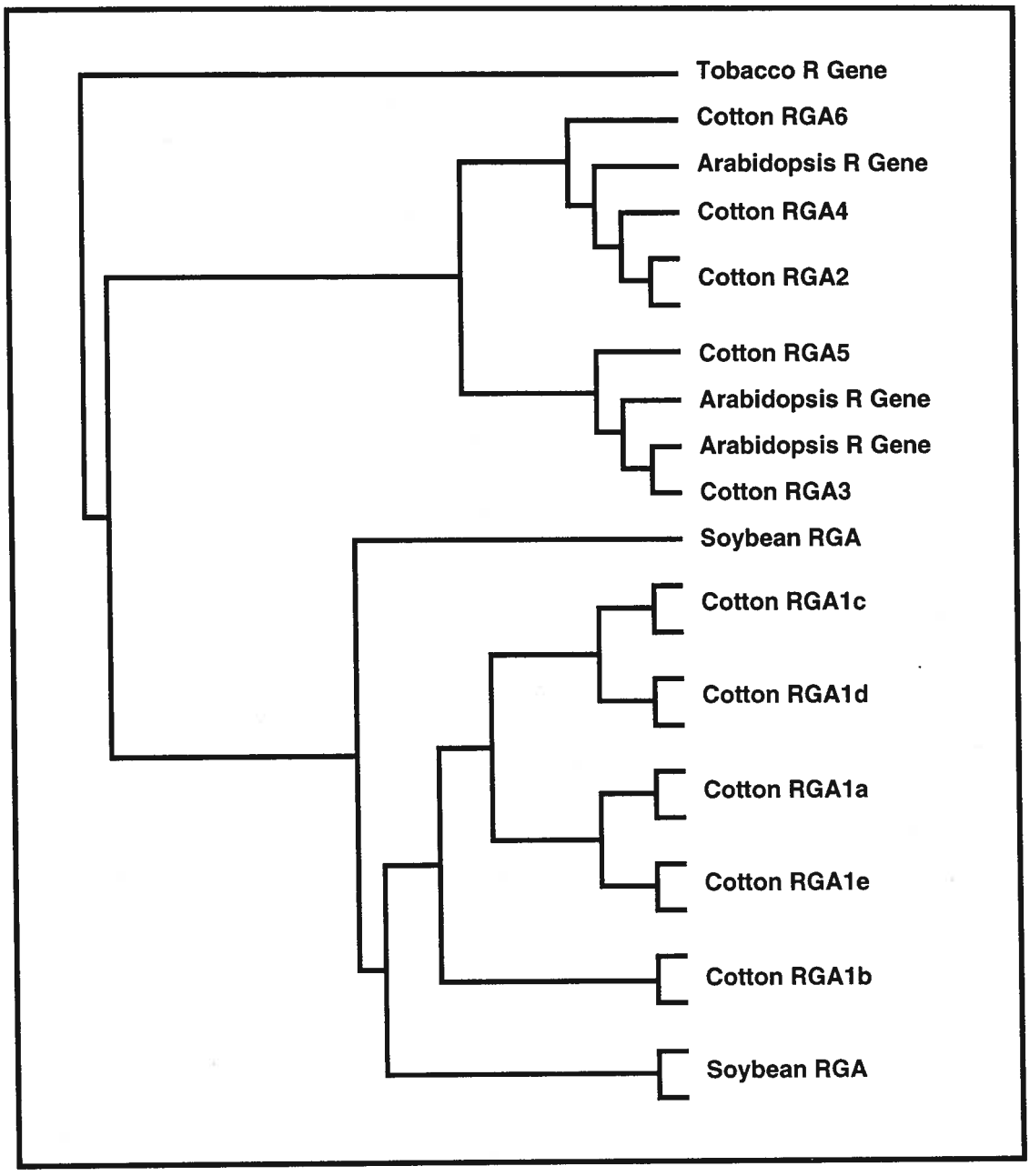

Figure 2. Family tree of cotton genes analogous to known disease resistance genes (cotton RGA families 1-6). Members of cotton RGA family 1 share similarities with RGAs from soybean. Members of cotton RGA families 2-6 share similarities with disease resistance genes from *Arabidopsis*.

The role of RGAs in creating disease resistant cultivars

The big challenge ahead is to determine which of these cotton RGAs represent true resistance genes, and what is the nature of the organism they provide protection against in cotton. We are initially targeting Verticillium wilt and Bacterial blight as examples of fungal and bacterial diseases of economic importance to the cotton industry. Cultivars of known disease response

such as Pima S-7 (*G. barbadense*, Wilt resistant, Blight susceptible) and CS50 (*G. hirsutum*, Wilt susceptible, Blight resistant) are being employed. These cultivars allow us to compare the DNA fragment patterns of the RGAs in each plant and to relate presence or absence of a particular DNA fragment with disease response.

We have established the presence of RGAs in both *G. hirsutum* and *G. barbadense* and subtle differences have been detected that may account for the variation detected in disease resistance levels between the cotton species. Our expectation is to find a RGA linked with Verticillium wilt resistance in Pima S-7, and a RGA linked with Bacterial blight resistance in CS 50. Linked RGAs could then be used as DNA markers to enable selection of plants carrying genes for resistance to these diseases.

Knowledge and understanding of the process of pathogen recognition and defence by plants holds great promise for the development of better disease control in cotton. If we can identify the resistance genes responsible for recognition of the organisms causing such diseases as Verticillium wilt, Fusarium wilt or Bacterial blight, we can ensure that all of these genes are included when breeding improved, more resistant cotton varieties for Australian conditions.

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Reference

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